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A Cross-sectional Study of Iron Deficiency Anemia and Hemoglobinopathy Carriers among School Children in Two Different Cities in KSA

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Abstract

BACKGROUND: Iron deficiency anemia (IDA) is the most common cause of anemia in the Kingdom of Saudi Arabia. Thalassemia and sickle cell disease are major concerns in several nations, including KSA.

AIM: This study sought to investigate the prevalence of IDA, sickle cell trait (SCT), and B thalassemia trait (BTT) among school children in Al-Khobar and Makkah, Saudi Arabia.

METHODS: A multistage random sample of 570 school children aged 12–18 years from both cities were educated about these illnesses and tested for CBC, serum iron, TIBC, reticulocytosis, and sickling. In some circumstances, hemoglobin electrophoresis and HPLC were used.

RESULTS: Overall, 13% of people had IDA, 3.2% had SCT, and 2.5% had BTT. The hematological parameters of students with IDA and BTT differed significantly from those of normal students in each city, but not between normal students and those with SCT. There were no significant differences between the sexes for all blood parameters except iron and TIBC.

CONCLUSION: Strengthening public health education, dietary initiatives, and school-based hemoglobinopathy carrier screening is proposed to help detect anemia early.

Introduction

In many regions of the world, iron deficiency anemia (IDA) is one of the most prevalent nutritional illnesses [1]. In Saudi Arabia, it disproportionately affects women and children [2]. Hemoglobinopathies (hemoglobin disorders) are the most prevalent single gene disorders in the world's population [3] and are characterized by the production of structurally abnormal hemoglobin variants such as sickle cell disease (SCD) and sickle cell trait (SCT) or by a decrease in the synthesis of structurally normal globin (B-thalassemia) [4].

Saudi Arabia is well-known for having a high rate of genetic blood diseases [5]. Over the last decade, the premarital screening program has undertaken surveys for hemoglobinopathies in several KSA regions. Numerous studies have revealed relatively high rates of B-thalassemia carriers in the Eastern area (19.5%), Qunfudah (15.8%), Jizan (7.8%), and Northern border region (7.8%) (7.25%). Moderate frequencies were reported in Al-Ahsa (3.4%), Makkah (2.7%), Riyadh (2.01%), and Jeddah (2.01%) (1.23%). Other locations, such as Taif (0.43%), Madinah (0.41%), Tabuk (0.30%), Qassim (0.16%), Jouf (0.07%), and Hail (0.07%), recorded lesser frequencies (0.02%) [5], [6]. Numerous factors may contribute to the high prevalence of hemoglobinopathies in the KSA, including demographic factors such as rapid population growth and an increase in the number of Asian immigrants; also certain cultural, traditional, and religious characteristics, such as a high rate of consanguineous marriages (over 55%), particularly first cousin unions, early marriage, large family size, and advanced paternal and maternal ages [7], [8]; in addition, genetic variety is evident among Saudi people as a result of mingling with other nations, particularly those from hemoglobinopathies-endemic areas in East and South Asia [9].

Hemoglobinopathies management faces numerous challenges at various levels: At the national level due to a lack of resources, the presence of other competing priorities such as communicable and non-communicable diseases, an insufficient number of trained health professionals in this field with low genetic literacy in the health sector, and an absence of data on the true magnitude and economic burden of hemoglobinopathies; and at the community level due to a lack of public awareness, and finally the cultural concern of affected families being stigmatized within their society

has a substantial psychosocial and emotional impact on patients and their families [10], [11], [12].

Saudi health authorities' attention to hemoglobinopathies has focused on neonatal screening for SCD in order to initiate early treatment and preventive interventions prior to the onset of clinical symptoms. It is, however, only useful when there is appropriate genetic counseling and parental education, as well as adequate primary and follow-up care for those affected to reduce morbidity and mortality, additionally on premarital screening for carriers of B-thalassemia and SCD to assist couples who are at high risk of having an affected baby to make an informed reproductive decision and choice. It should, however, be done in a way that respects the population's religious, traditional, and cultural perspectives while also dealing with social and ethical dilemmas, government policies, and the attitudes of the couples themselves. Such screening efforts will be ineffective in decreasing the burden of hemoglobinopathies if options are not made available to carrier couples [13], [14]. If carrier couples are not offered options, such screening initiatives will be ineffective at reducing the burden of hemoglobinopathies [10]. One possibility to consider is instituting screening during the school years [15].

We believe that adolescent school students represent an excellent opportunity for screening and counseling for genetic blood disorders because they are aware of the potential risk of having affected children at this age, have time to process and appreciate the information before choosing a partner, and thus are less likely to marry another carrier in order to reduce the risk of these diseases in their future families. Additionally, such screening may assist raise awareness and educate kids and the linked demographic group, which includes parents, teachers, friends, siblings, and employees, about these diseases.

The purpose of this study was to determine the prevalence of and raise awareness about IDA and hemoglobinopathies, particularly SCT and TT, among adolescent school students in Al-Khobar and Makkah cities, Saudi Arabia, to emphasize the importance of setting priorities for community-based prevention programs and more efficient health service planning.

Methods

Study area and population

An institutional-based cross-sectional descriptive study was conducted between between November 2018 and April 2020 during the school months among preparatory and secondary school children aged 12–18 years in 20 public schools (10 for males and 10 for females) in Al-Khobar, as a representative of the Eastern area, and in Makkah, as a representative of the Western area, Saudi Arabia.

Sample size

The sample size was determined using the Statcalc from Epi Info version 6.04 (Centers for Disease Control and Prevention, Atlanta, USA) with a sample size of 454 at 95% level of confidence, 2% margin of error, 80% power, and an expected prevalence of hemoglobinopathies 5%. We tried to compensate for possible losses and improve the validity of results, by increasing the final sample size to 600 students. After selection, we excluded 23 students due to refusal of sampling, missing blood reports and technical laboratory errors and another seven students were excluded who have been diagnosed with thalassemia major, sickle cell anemia or other recognized hematological diseases. Other non-Saudi nationalities were excluded, and accordingly the final sample size enrolled in the study was 570 students.

Sampling method

The multistage sampling method was used in each city, whereby schools were stratified according to general education departments into east, west, north, south, and middle with sub-stratification into two strata, preparatory and secondary schools. Schools were chosen randomly from each stratum to represent male and female students. Random selection of students was considered proportional to their size in the study schools. In each school, sampling frame was prepared from the student roster in each grade and students were selected from each grade by simple random sampling method. A total of 294 students were chosen randomly from 10 schools in Al-Khobar and a total of 276 students were chosen randomly from ten schools in Makkah (with equal number of male and female schools in each city).

Procedures

Before blood collection, instructors and students were oriented through lectures, the distribution of posters and booklets, and the display of banners, in collaboration with the management in each school. Each student had a 5 ml venous blood sample drawn; first, 3 ml blood samples were drawn into an EDTA tube to analyze complete blood count (CBC), reticulocytic count, and sickling test for all students, as well as high performance liquid chromatography (HPLC) and hemoglobin electrophoresis in suspected cases. For all students, the remaining 2 ml blood samples were clotted to quantify serum iron and total iron binding capacity (TIBC) using a calorimetric technique. A Cell-Dyn 3700 was used to determine different CBC parameters, Red cell indices and RDW (red-cell distribution width) red cell morphology (target, sickle, pencil, hypochromia). In addition, reticulocytic count (RTX) was performed on all pupils using Brilliant cresyl blue stain to distinguish between types of hemolytic anemia and other types of anemia [16]. A sickling test was also performed on all students to detect the presence of Hbs in SCD or SCT.

All samples with anemia (Hb 11 g/dL and/or low MCV 68 fl) and/or high (RTX) and/or positive sickling test were electrophoretically separated of (Hb-EPS) on cellulose acetate paper. The aberrant Hb bands are detected using Hb-EPS (HbS, F and A2). We perform another confirmatory test using microcolumn chromatography to discover the real amount of HbA2 [17].

- The presence of low red cell indices (MCV 68 fl, MCH 23 pg), RBC morphology [as pencil cells], low RTX count (0.5%), high RDW (>17%), and iron saturation (15%) was used to diagnose IDA.

- TT was identified in instances with atypical RBC shape (as Ovalocytosis and Target), decreased blood indices, low RDW (13.5%), high RTX (>2.5%), raised HbA2 (>3.5%), and high iron saturation (>46.0 ug/dl). The true level of HbA2 (was determined using column chromatography.

- SCT was verified by (Hb-EPS) and (HPLC) with near normal Hb, blood indices, and RTX.

Statistical analysis

Statistical analysis was carried out using the SPSS computer package version 25.0 (SPSS Inc., Chicago, IL, USA). For descriptive statistics: The mean ± SD were used for quantitative variables while the number and percentage were used for qualitative variables. Fischer's exact test (FET) was used to assess the differences in frequency of qualitative variables while Mann–Whitney U-test or Kruskal–Wallis test were applied to assess the differences in means of quantitative variables with Tukey HSD *post hoc* correction to indicate which groups were significantly different from which others. The statistical methods were verified, assuming a significant level of $p < 0.05$ and a highly significant level of $p < 0.001$.

Results

A total of 570 Saudi students were enrolled in this study; 294 (51.6%) from Al-Khobar (141 from preparatory and 153 from secondary schools) while the other 276 (48.4%) were from Makkah (132 from preparatory and 144 from secondary schools). Their mean age was 14.7 ± 3.6 years ranged from 12 to 18 years and 305 students (53.5%) were males (161 from Al-Khobar and 144 from Makkah) with no significant difference between both cities regarding age ($p = 0.104$) and gender ($p = 0.557$).

A total of 106 students (18.6%, 58 males and 48 females) exhibit either IDA or hemoglobinopathies where 60 students were from Al-Khobar and 46 from Makkah. The overall prevalence of IDA was 74 cases (13%) with 39 from Al-Khobar and 35 from Makkah, and 40 males and 34 females. The overall prevalence of SCT was 18 cases (3.2%) with 11 from Al-Khobar and 7 from Makkah, and ten males and eight females. The overall prevalence of βTT was 14 cases (2.5%) with 10 from Al-Khobar and 4 from Makkah, and eight males and six females. There were no significant differences between both cities as regard prevalence of IDA (P= 0.901), SCT ($p = 0.478$), or βTT ($p = 0.177$) (Figure 1).

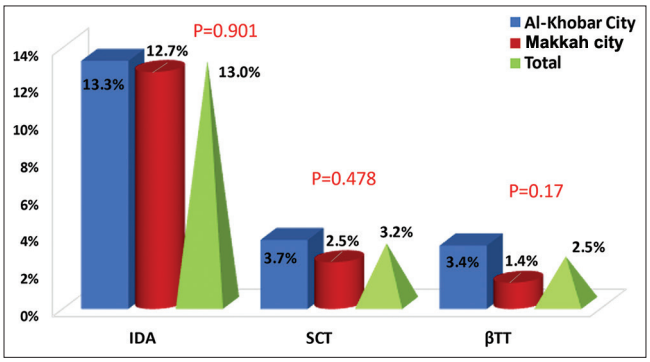

Figure 1: Prevalence of different types of anemia among school children in Al-Kobar and Makkah

The hematological characteristics between the normal students and those with IDA or carriers of hemoglobinopathies (SCT/βTT) in both cities were shown in (Tables 1 and 2). Within each city, the mean values of Hb, HCT, MCV, MCH, and RTX were significantly lower in students with IDA and βTT compared with normal students except the mean RTX was significantly higher in students with βTT compared with normal students. However, no significance exists in all mean hematological parameters between normal students and those with SCT in both cities.

Among students with IDA or carriers of hemoglobinopathies (SCT/βTT), no significant differences were detected (P>0.05 in all) when comparing the mean hematological values among school children in both cities (Table 3).

Table 1: Mean hematological values among school children in Al-Khobar

Parameters	Al-Khobar (n = 294)				p-value
	IDA (n = 39)	SCT (n = 11)	βTT (n = 10)	Normal (n = 234)	
Hb (g/dl)	10.1 ± 1.9	12.5 ± 1.6	10.3 ± 1.5	13.0 ± 0.9	< 0.001[1, 2*]
HCT (%)	32.4 ± 2.7	37.8 ± 5.4	31.5 ± 2.4	40.5 ± 4.8	< 0.001[1, 2*]
MCV (fl)	67.4 ± 7.1	84.5 ± 5.6	68.3 ± 6.5	82.4 ± 4.9	< 0.001[1, 2*]
MCH (pg)	18.5 ± 4.2	26.9 ± 4.0	21.4 ± 3.8	27.2 ± 3.6	< 0.001[1, 2*]
MCHC (g/dl)	32.6 ± 3.5	33.3 ± 4.0	31.7 ± 2.3	33.6 ± 3.0	0.087
RBCs ($x10^6$/L)	4.6 ± 1.2	4.7 ± 1.3	5.3 ± 0.8	4.9 ± 0.9	0.131
WBCs ($x\ 10^3$/L)	7.9 ± 2.4	9.0 ± 1.8	8.6 ± 2.5	8.2 ± 2.1	0.452
Platelet ($x\ 10^3$/L)	414.7 ± 64.3	381.1 ± 40.5	392.6 ± 34.9	406.4 ± 38.4	0.097
RTX (%)	0.92 ± 0.73	1.81 ± 0.92	2.28 ± 1.15	1.42 ± 1.06	0.001[1, 2*]

Values present as mean ± SD & analyzed by Kruskal-Wallis test. [1]Significance between normal and IDA values, [2] Significance between normal and βTT values. No significance between normal and SCT values. *:Significant.

Table 2: Mean hematological values among school children in Makkah

Parameters	Makkah (n = 276)				p-value
	IDA (n = 35)	SCT (n = 7)	βTT (n = 4)	Normal (n = 230)	
Hb (g/dl)	9.6 ± 1.8	13.3 ± 1.1	9.8 ± 1.7	13.2 ± 1.2	<0.001[1, 2]*
HCT (%)	29.8 ± 2.3	39.7 ± 4.1	30.6 ± 2.6	40.3 ± 4.5	<0.001[1, 2]*
MCV (fl)	70.2 ± 7.3	84.9 ± 4.9	69.0 ± 6.6	83.1 ± 5.8	<0.001[1, 2]*
MCH (pg)	21.6 ± 3.9	27.0 ± 2.9	23.1 ± 2.5	28.4 ± 3.7	<0.001[1, 2]*
MCHC (g/dl)	32.3 ± 3.4	32.5 ± 3.6	31.9 ± 3.9	34.0 ± 4.1	0.074
RBCs (× 10^6/L)	4.9 ± 1.2	4.7 ± 1.5	5.5 ± 1.6	5.1 ± 1.1	0.516
WBCs (×10^3/L)	8.2 ± 2.5	8.8 ± 2.7	7.8 ± 2.2	7.5 ± 2.9	0.379
Platelet (× 10^3/L)	370.6 ± 49.9	360.1 ± 54.6	318.9 ± 51.8	349.3 ± 50.7	0.067
RTX (%)	0.58 ± 0.42	1.66 ± 0.78	2.71 ± 0.63	1.47 ± 0.81	<0.001[1, 2]*

Values present as mean ± SD & analyzed by Kruskal-Wallis test. [1]Significance between normal and IDA values. [2] Significance between normal and βTT values. No significance between normal and SCT values. *:Significant.

Students with IDA or carriers of hemoglobinopathies (SCT/βTT) showed Hb, RBCs, and HbA2 lower than normal. No statistically significant differences were observed between both genders for any of the hematological characteristics examined except among students with IDA where iron was significantly decreased in females (10.8 ± 2.7 vs. 13.2 ± 3.4; p = 0.001) and TIBC was significantly decreased in males (392.4 ± 52 vs. 419.7 ± 47; p = 0.021) (Table 4).

Verifying quality

Aside from meticulously following manufacturers' instructions and normal operating procedures, all reagents were tested for expiration dates to assure data quality. Each student's name, age, gender, school grade, and lab results were documented on standard report formats.

Ethical concern

Both cities' health and education ministries approved the project, as did each school's principal. Parents signed informed consent forms and pupils gave oral approval before sampling. Throughout the trial, a unique code number protected privacy and confidentiality. Students with anemia/hemoglobinopathies were sent to health-care consultation.

Discussion

IDA is a worldwide hematological and public health issue. Because most prior studies focused on young children and pregnant females or girls, investigations on its incidence among adolescent school pupils in KSA are largely missing [18].

Adolescents (13%) had IDA, which could be due to increased iron demand during puberty, menstrual losses, low dietary iron intake, poor dietary habits, nutritional deficiencies, and poor iron absorption [19]. Sedentism, fast-food consumption, and preference for indoor activities have recently been linked to IDA [20]. However, the prevalence was lower than that reported in previous studies among school students conducted in different regions of KSA, which ranged from (16.1%) among primary school girls in Riyadh [21] to a recent prevalence of (16.7%) in male and (34.2%) in female adolescents aged 13–18 years [22], (20.5%) among a sample of 800 Saudi school children in Jeddah [23], and up to (24.8%) in different areas of the country with highest prevalence (16.5%) [24]. The lower incidence in our study could be due to widespread iron fortification of wheat flour. Because IDA levels among school children are still worrying, public health officials must focus on boosting nutrition and educational initiatives.

Similar prevalence (11.18%) among school children in Jordan [25] and Egypt [26], but higher prevalence (17.9%) among Arab migrating nomad school children in Iran [27], (20.35%) among adolescents intermediate school pupils in Iraq [28], and (23%) among Kuwaiti student [29]. The higher results in Iraq may be due to low socioeconomic status and the economic embargo imposed on Iraq for the last decade, whereas in Kuwait, it may be due to bad dietary habits (delivery meal) and excessive calories consumed, as seen by the estimated 40% obesity rate [30].

Hemoglobinopathies are hereditary illnesses with varied prevalence rates and molecular

Table 3: Mean hematological values associated with (IDA/SCT/βTT) among school children in both cities

Parameters.	IDA (n = 74)		SCT (n = 18)		βTT (n = 14)	
(Positive results)	Al-Khobar (n = 39)	Makkah (n = 35)	Al-Khobar (n = 11)	Makkah (n = 7)	Al-Khobar (n = 10)	Makkah (n = 4)
Hb (g/dl)	10.1 ± 1.9	9.6 ± 1.8	12.5 ± 1.6	13.3 ± 1.1	10.3 ± 1.5	9.8 ± 1.7
HCT (%)	32.4 ± 2.7	29.8 ± 2.3	37.8 ± 5.4	39.7 ± 4.1	31.5 ± 2.4	30.6 ± 2.6
MCV (fl)	67.4 ± 7.1	70.2 ± 7.3	84.5 ± 5.6	84.9 ± 4.9	68.3 ± 6.5	69.0 ± 6.6
MCH (pg)	18.5 ± 4.2	21.6 ± 3.9	26.9 ± 4.0	27.0 ± 2.9	21.4 ± 3.8	23.1 ± 2.5
MCHC (g/dl)	32.6 ± 3.5	32.3 ± 3.4	33.3 ± 4.0	32.5 ± 3.6	31.7 ± 2.3	31.9 ± 3.9
RBCs (x 10^6/L)	4.6 ± 1.2	4.9 ± 1.2	4.7 ± 1.3	4.7 ± 1.5	5.3 ± 0.8	5.5 ± 1.6
WBCs(x 10^3/L)	7.9 ± 2.4	8.2 ± 2.5	9.0 ± 1.8	8.8 ± 2.7	8.6 ± 2.5	7.8 ± 2.2
Platelet (x 10^3/L)	414.7 ± 64.3	370.6 ± 49.9	381.1 ± 40.5	360.1 ± 54.6	392.6 ± 34.9	318.9 ± 51.8
RTX (%)	0.92 ± 0.73	0.58 ± 0.42	1.81 ± 0.92	1.66 ± 0.78	2.28 ± 1.15	2.71 ± 0.63
RDW (%)	17.2 ± 2.6	16.6 ± 3.1	13.8 ± 2.4	13.4 ± 2.1	12.8 ± 3.0	12.5 ± 2.6
HbA2 (%)	1.55 ± 0.71	1.71 ± 0.43	2.35 ± 1.27	2.29 ± 1.45	5.62 ± 1.13	5.44 ± 1.27
Iron (ug/dl)	13.24 ± 3.5	11.9 ± 4.4	42.8 ± 3.2	40.9 ± 2.9	45.4 ± 3.0	43.8 ± 2.7
TIBC (ug/dl)	411.2 ± 52	434.6 ± 55	281.6 ± 47	303.5 ± 60	234.0 ± 36	246.5 ± 44
Hb-EPS	Predominant HbA, Normal HbA2	Predominant HbA, Normal HbA2	HbA, HbS, Normal Hb A2	HbA, HbS, Normal HbA2	HbA, HbF, High HbA2	HbA, HbF, High HbA2
Sickle test	-	-	positive	positive	-	-

Table 4: Mean hematological values associated with (IDA/SCT/βTT) among school children in both genders

Parameters.	IDA (n = 74)		SCT (n = 18)		βTT (n = 14)	
(Positive results)	Male (n = 40)	Female (n = 34)	Male (n = 10)	Female (n = 8)	Male (n = 8)	Female (n = 6)
Hb (g/dl)	9.4 ± 1.6	8.8 ± 1.9	12.8 ± 1.4	12.2 ± 1.3	10.6 ± 1.8	10.2 ± 1.6
HCT (%)	28.3 ± 2.7	27.9 ± 2.9	36.4 ± 1.1	39.0 ± 1.3	38.2 ± 0.7	38.3 ± 0.9
MCV (fl)	53.5 ± 11.7	56.2 ± 12.8	84.4 ± 3.2	85.2 ± 3.5	60.9 ± 9.4	61.5 ± 11.0
MCH (pg)	17.7 ± 5.4	18.3 ± 4.6	26.0 ± 3.8	27.8 ± 2.9	21.4 ± 2.9	21.8 ± 3.0
MCHC (g/dl)	33.1 ± 3.8	35.2 ± 2.4	32.7 ± 4.6	32.2 ± 4.0	31.8 ± 2.0	31.0 ± 1.7
RBCs (× 10^6/L)	4.4 ± 1.8	4.3 ± 1.9	4.5 ± 2.4	4.6 ± 2.1	4.5 ± 1.5	4.5 ± 1.8
WBCs (× 10^3/L)	7.3 ± 3.3	7.6 ± 3.0	10.0 ± 3.2	10.5 ± 3.5	7.6 ± 2.5	7.8 ± 2.8
Platelet (× 10^3/L)	449.8 ± 65	472.3 ± 64	332.8 ± 39	379.1 ± 50	318.6 ± 41	330.9 ± 37
RTX (%)	0.56 ± 0.26	0.49 ± 0.34	2.21 ± 0.56	2.10 ± 0.44	2.63 ± 0.82	2.77 ± 0.61
RDW (%)	17.9 ± 2.5	18.3 ± 2.7	14.1 ± 2.2	13.7 ± 2.4	12.6 ± 2.7	12.3 ± 2.6
HbA2 (%)	1.5 ± 0.9	1.7 ± 0.8	2.28 ± 1.25	2.33 ± 1.34	5.5 ± 1.2	5.4 ± 1.4
Iron (ug/dl)	13.2 ± 3.4[1]	10.8 ± 2.7[1]	42.2 ± 4.3	41.6 ± 4.9	47.5 ± 4.4	46.7 ± 5.0
TIBC (ug/dl)	392.4 ± 52[1]	419.7 ± 47[1]	308.1 ± 51	298.4 ± 45	218.6 ± 32	224.8 ± 36
Hb-EPS	Predominant HbA, Normal HbA2	Predominant HbA, Normal HbA2	HbA, HbS, Normal HbA2	HbA, HbS, Normal HbA2	HbA, HbF, High HbA2	HbA, HbF, High HbA2
Sickle test	-	-	positive	positive	-	-

Values present as mean ± SD & analyzed by Mann-Whitney *U*-test. [1]: Statistically significant difference ($p < 0.05$).

characteristics [13]. Our study indicated an overall frequency of B-thalassemia carriers among school students (2.5%), worldwide; the rate of B-thalassemia carriers varies. There were also reports of (1.06%) among 3571 high school Turkish students aged 12–22 years [31], (2.9%) among 5685 Bahraini students in the 11th grade aged 16–17 years [32], (3.04%) among school children aged 6–15 years in north-eastern Badia region of Jordan[22], (3.22%) among 9–12 year old's in Pakis [33].The disparities may be due to variances in age groups investigated, socio-cultural differences between areas, or sample sizes.

In terms of SCT, our study found a prevalence of (3.2%), although Ashour (2004) showed a lower prevalence among primary school pupils in Makkah (1.71% in boys and 1.28% in girls) [34]. There were also reports of prevalence of (13.8%) among Bahraini students aged 16–17 [32], (6%) among Omani children aged 0–5 [35]. The discrepancy in prevalence of hemoglobinopathies among different countries may be explained by differences in study design, estimating methodology, and ethnicity.

Other types and mutations of hemoglobinopathies were not screened, students from private schools were not included, and the relative frequency in the overall population cannot be judged from these data because this study was done in a specific group of population that may not reflect the true prevalence.

Conclusions

New epidemiological data on IDA and hemoglobinopathies among teenage school students in KSA may be useful for genetic counseling and premarital screening. Updating the school curriculum, routine carrier screening among school children, updating medical and nursing college curriculums related to the practice of these disorders, and integrating community genetic services into primary health-care systems help planners and policy-makers with reliable information. More research is needed to assess the long-term effects of partner testing and premarital counseling in this age range.

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